IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant(s): Csöregi et al	
Application No.: 10/019,651	Group Art Unit: 1651
Filed: 7/8/2002	Examiner: Jon P. Weber
Title: BIOSENSOR	Confirmation No:
Attorney Docket No.: 50159-026	

DECLARATION UNDER 37 CFR § 1.132

The undersigned hereby declares as follows:

1. We are named inventors of the above-referenced application, and we are familiar with the application, including the claims thereof.

2. We understand that an Official Action has issued in this case in which the Examiner takes the position that the enzyme from grass pea is more selective and sensitive than the enzyme from *Aspergillus*. We further understand that the method of the present invention would be relevant if this method results in a different, improved result compared to said obtained using *Aspergillus*.

3. Tests have been conducted to demonstrate the effects obtained using amine oxidase from grass pea, and from *Aspergillus*, respectively, particularly in demonstrating presence of histamine using transducers.

The following comparison data are present, where amine oxidase from grass pea have been compared with amine oxidase from *Aspergillus niger*. The comparison happens to be related to different transducer systems, but still it is apparent that amine oxidase from *Aspergillus niger* provides a lower sensitivity than amine oxidase from grass pea (0.93 vs. 0.48), as well as the same high potential is not needed (+200 mV vs. +600 mV). High potentials will

lead to high background currents and a small selectivity due to disturbing signals related to electro chemical interferences. A system comprising HRP (horse radish peroxidase) will become quite different, but provides on the other hand for a high selectivity with regard to putrescine and cadaverine. Amine oxidase from *Aspergillus niger* can not be used for putrescine, but then a peroxidase from *Micrococcus rubens* is needed.

ENZYME	TRANSDUCER	E vs. Ag/AgCl (mV)	SELECTIVITY	LR (µM)	DL (µM)	S (mA/M)
AO grass pea HRP	Graphite	-50	His 100 % Put 147 % Cad 132 %	1-150 1-400 1-400	0.33 0.17 0.20	5.16 13.58 11.80
AO grass pea	Graphite	+200	His 100 % Put < 1 % Cad < 1 %	10-200 - -	2.2 - -	0.48 - -
AO A. niger	Pt paste	+600	His Put	0.17-20 0.06-200	n.a. n.a.	0.93 2.4

LR stands for Linear range
DL stands for Detection limit
S stands for Sensitivity

We hereby declare that all statements made herein of our own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

dated: 26/11 -2003 Elisabeth Csöregi

Elisabeth Csöregi

dated: 26/11 -2003 Mihaela Niculescu

Mihaela Niculescu

dated: 17/12 -2003

Ivo Frébort